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A review of lipid-based biomasses as feedstocks for biofuels production

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ABSTRACT

This review aims to provide up-to-date knowledge on existing feedstocks for biofuels production (mainly biodiesel) from lipid-based biomasses. The 1st generation of lipid-based feedstocks was edible plant oils, whilst other alternative feedstocks were discovered and reported as the 2nd generation feedstocks. The 2nd generation feedstocks that are summarized in this work include non-edible oils, waste vegetable oil, animal fats, industrial wastes and by-products, lipid derived from insects and microorganisms. The general strong points of the 2nd generation feedstocks are that they are inexpensive, of high productivity and typically do not compete ethically or economically with food crops (edible oils). However, all 2nd generation feedstocks usually have a high level of moisture and free fatty acids that cause an extremely negative effect on conventional biodiesel production process. Thus, this article provides basic information on the processing techniques that are capable of handling 2nd generation feedstocks as well.

Contents

1. Introduction

Petroleum resources play an important role as the major energy source worldwide, especially for transportation. However, the World's accessible petroleum resources, which are non-renewable, are gradually depleting and being projected to exhaust in the future. The biofuel synthesized from lipid-based biomasses is also

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a promising transportation fuel. The term "biofuels" in this article includes biodiesel that is chemically synthesized by transesterification and/or esterification reactions and other biofuels that are thermo-chemically produced via pyrolysis, catalytic cracking and supercritical technique. Biodiesel quality, which influences the in-use performance of diesel engines, is specified by international standards, such as EN14214 and ASTM D6751-08, and consequentially has gained more interest than other biofuels that are produced from lipid-based biomasses. Thus, the term "biofuels" in this article mostly refers to biodiesel. The alternative biofuels that are thermo-chemically produced from lipid-based biomasses normally fail to meet the designated 96.5% ester content in the International standard of biodiesel (EN14214), but their fuel properties make them of interest as alternative transportation fuels [1]. Please keep in mind that the term "biofuels" in this article excludes other chemicals synthesized from sugar-based biomasses, such as methanol, ethanol and dimethyl ether.

The content of this review begins with a brief overview on plant oils, especially edible oils as the 1st generation of lipid-based biomass feedstocks. Then, non-edible plant oils, animal fats, industrial wastes and by-products are introduced as the 2nd generation feedstocks. Next, some insects and oleaginous (oil rich) microorganisms are also reported as another 2nd generation feedstock. Finally, recommendations and perspectives of biofuel processing for the 2nd generation lipid-based feedstocks are provided at the end of this article.

2. Lipid-based biomasses from oil plants

2.1. Edible oil plants

Lipid-based biomasses derived from edible oil plants, namely cooking oils, are classified into the two groups of (i) saturated oils, such as coconut and palm kernel oils, and (ii) the unsaturated oils, such as canola, soybean and sunflower oils. The typical fatty acids contained in the edible oils and fat are lauric acid (C12:0), myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1), linoleic acid and linolenic acid (C18:3). Note that the code (CX:Y) refers to numbers of carbon atoms (X) and C=C double bonds (Y) in the fatty acid molecule. The definitive works on edible oil compositions, properties, characteristics, processing and application are available elsewhere [2,3].

Edible oils are the 1st generation of biofuel production from lipid-based biomasses because of their availability. From the biofuel production point of view, the degree of saturation and fatty acid chain length slightly affect the production pathway and properties of the resulting products [4]. However, the most important issue for using lipid-based biomasses as biofuel feedstocks is the oil productivity, which is somewhat reflected in its price, as illustrated in Table 1.

Table 1
The lipid content and productivity of general edible oil plants [82,83].

Oil	Lipid content (%wt)	Oil yield (kg/ha/yr)	Price[a] (US$/kg)
Oilseeds			
Canola	40–45	590.7–663.8	1.23
Corn	3–6	241.9–438.8	1.55
Cottonseed	18–20	208.1–236.3	1.72
Peanut	45–50	1260.1–1400.8	2.26
Soybean	18–20	450–506.3	1.14
Sunflower	35–45	517.6–663.8	1.52
Tree fruits and kernels			
Coconut	65–68	731.3–978.8	0.89
Olive	15–35	101.3–292.5	3.58
Palm	45–50	3004–5006	0.82
Palm kernel	45–50	300.4–500.7	0.88

[a] Refers to the average price between Aug 2012–Feb 2013 (from www.indexmundi.com).

Oil palm has a high potential as a biofuel feedstock due to having the highest productivity. However, it only grows well in tropical climates within a narrow band of 5° north and south of the equator. The ideal growing conditions for oil palm are a moderate temperature (25–33 °C), over 2000 h per annum of sunshine, and rainfall of over 200 cm per year but spread evenly throughout the year. Thus, oil palm plantations are only suitable for biofuel feedstocks in some particular countries, and in addition it has a climate risk. The oil palm is a perennial plant that requires a few years before being partially harvestable and two more years before being fully harvestable. Although oil palm farming requires a large amount of water, the monsoon rain in the tropical climate can cause flooding that immediately damages young oil palm trees and adversely impacts upon the crop yield in the future. The top five palm oil producing countries in 2011, as reported in the Index Mundi website, were Indonesia, Malaysia, Thailand, Colombia and Nigeria, respectively.

Coconut trees also have high oil productivity, but also require a large amount of rainfall (130–230 cm per year) and plenty of sunshine, somewhat similar to the oil palm tree. Like oil palm trees, coconut trees require 4.5 years growth before they can be harvested, during which time they can be harmed by either rain storms or other climate risks in the tropical climate. The principal coconut oil producers in 2011, as reported in the Index Mundi website, were the Philippines, Indonesia, India, Vietnam and Mexico, respectively.

Olive oil is mostly produced (up to 80% of the worldwide level), in the EU, with the main countries being Spain, Italy and Greece, and is principally consumed as a food seasoning. Because olive oil has a high price, the biofuel derived from it is likely to be more expensive than that derived from other oil plants. On the other hand, sunflower seeds have the potential to be used as a biofuel feedstock in the EU rather than olive trees. For instance, over 80% of sunflower oil worldwide is produced in the Ukraine, Russian Federation and other European countries.

In 2011, more than 80% of the global peanut oil production was produced in China and India. However, as with olive oil, its price is not economically competitive when compared to that of other plant oils. Despite the low lipid content of soybean, it has the highest total production of over 40 million tons worldwide in 2011. The soybean is also an annual crop and is farmed around the world including in Asia, Europe, and North and South America. The top five soybean oil producing countries in 2011, as reported in the Index Mundi website, were China, United States, Argentina, Brazil and India.

Nevertheless, the lipid-based biomasses derived from edible oil are currently only sufficient to meet the available household and industrial demands worldwide. Therefore, if used to supply biofuel production, the price of those biomasses will increase. For example, the price of crude palm oil in Thailand increased from 0.80 US$/kg to 1.23 US$/kg after the government announced that 5% (v/v) of biodiesel will be added to diesel fuel nationwide in 2015. Therefore, non-edible oils have become of interest as 2nd generation feedstocks since they are attractive resources to avoid the economic and ethical competition between the food and fuel industries.

2.2. Non-edible oil plants

Some plant families, such as *Brassicaceae* and *Euphorbiaceae*, have a high oil content in their mature seed, but are not

considered as suitable for human food or animal feed due to the significantly high free fatty acid (FFA) content, differences in their fatty acid profile or the presence of toxic substances [5]. The development of non-edible oil plants, especially for degraded or semi-arid lands that are inappropriate for food crops, has been introduced to provide an option for biofuel industries. The high potential non-edible oil crops and plants for biofuels production are shown in Table 2.

For Asian countries, many non-edible oil plants have been proposed as suitable feedstocks; with *Jatropha curcas* (Physic nut), *Pongamic pinnata* (Karanja tree) and *Azadirachta indica* (Neem) in India [6], and *Euphorbia lathyris* (Caper spurge), *Sapium sebiferum* (Chinese tallow tree) and *J. curcas* in the People's Republic of China [7]. From the Web of Knowledge, a total 149 of articles on the topic of non-edible oils were published during 2005–2011, of which 88 came from India, suggesting the potential importance India places on using non-edible oil plants as biofuel feedstocks. On the other hand, research into non-edible plant oil cultivation is largely replaced by that into different sources in other continents. For instance, in the USA and the EU, research on lipid-based feedstocks focuses on oleaginous microorganisms [8–10].

The fatty acid profile of some non-edible oils (Table 3) is similar to that of some edible oils, such as *J. curcas* and *P. pinnata* oils that have a principal fatty acid profile that is broadly similar to that of soybean and peanut oils, respectively. In contrast, some non-edible oils obtained from the *Brassicaceae* family have a high amount (up to 40% (w/w)) of erucic acid (C22:1) which is not found in any edible oils. Although the physical and chemical properties of biodiesel depend on the fatty acid profile of the feedstocks, there is no key fatty acid that assigns all the important fuel properties [11,12]. In other words, the fuel properties of biodiesel could not fail the International standard range due to the changes in any one given fatty acid in the profile of different feedstocks.

When comparing the oil productivity of non-edible oil plants to edible oil plants, the non-edible oil plants clearly have a lower oil productivity than edible oil plants, which is due to a few reasons. Firstly, the non-edible oil plants have not yet been subject to the same selection (cultivation and selective breeding technologies) and domestication as the edible oil plants. The development of plantation technologies could enhance the oil productivity of the non-edible oil plants. Secondly, the non-edible oil plants have typically been grown in degraded lands that have a lower nutrient and water content and so impinge on optimal photosynthesis and resource allocation to lipid production. However, the growth of non-edible plants on arable areas with excessive fertilizer for use as biofuel feedstocks would likely decrease their advantage on price and non-conflicting land usage. With respect to improving the productivity and/or to modify the fatty acid profile, the vibrant and dynamic nature of research on the metabolic engineering of triglyceride transiting in oilseed plants has been reviewed elsewhere [13].

Table 2
The lipid content and productivity selected non-edible oil plants.

Oil seed	% Lipid content in dry biomass (w/w)	Oil productivity (kg/ha/yr)	% FFA (w/w)
***Brassicaceae* Family**			
Ethiopian or Abyssinian mustard (*Brassica carinata*) [84–87]	26.5–36.7	1280	0.4–0.6
Rapeseed (*Brassica napus* L.) [84,88]	40.0–47.1	1100–1780	N/R
Wild mustard (*Brassica juncea*) [88,89]	37.9	1750–2000	0.7–1.1
***Euphorbiaceae* Family**			
Physic nut (*Jatropha curcas*) [90,91]	30–39	~1500	1.9–14.9
Castor (*Ricinus communis*) [92]	40.8–49.8	259–754	0.4–3.4
Caper Spurge (*Euphorbia lathyris*)[7,93]	43.3–50.0	645–900	12.8
Other Families			
Tobacco (*Nicotiana tabacum*) [94,95]	35–49	252–1050	1.7–6.2
Karanja tree (*Pongamic pinnata*) [96]	25–50	320–640	8.3–20.0
Rubber tree (Hevea bresiliensis) [97]	35–40	160–640	17.0
Neem tree (*Azadirachta indica*) [98]	20–30	320–480	> 20.0
Chinese tallow tree (*Sapium sebiferum*) [7]	12–29	740–1850	2.35

N/R is not reported.

Table 3
Fatty acid profiles of selected non-edible oil plants.

Fatty acid	Seed						
	Wild mustard (*B. junces*)	Physic nut (*J. curcas*)	Tobacco (*N. tabacum*)	Castor oil (*R. communis*)	Karanja tree (*P. pinnata*)	Rubber tree (*H. resiliensis*)	Neem tree (*A. indica*)
Lauric acid (C12:0)		0.1					
Myristic acid (C14:0)	0.1	0.1–0.2					1.0
Palmitic acid (C16:0)	2.6–3.6	13.6–15.6	6.6	0.8–1.5	11.0	10.0	18.0
Stearic acid (C18:0)	0.9–1.1	7.4–4.2	3.1	0.8–2.0	6.8	8.7	14.0
Oleic acid (C18:1)	7.8–13.9	37.6–44.7	22.0	3.6	49.0	25.0	46.0
Linoleic acid (C18:2)	147.2–21.5	31.4–43.2	66.0	3.5–6.8	19.0	40.0	18.0
Linolenic acid (C18:3)	13.0–13.4	0.2–0.3	1.0			16.0	
Ricinoleic acid (C18:1:1OH)				82.0–95.0			
Arachidic acid (C20:0)	0.8		0.3			4.1	
Gaoleic acid (C20:1)	5.3–8.7				2.4		3.0
Behenic acid (C22:0)	1.5		0.4		5.3		
Erucic acid (C22:1)	33.5–45.7						

3. Lipid-based biomasses from wastes and by-products

3.1. Waste vegetable oil

Waste vegetable oils, such as spent frying oil that is not suitable for further use, are an alternative feedstock to edible oils for biofuel production [14,15]. Over the past decade, waste vegetable oils have been mixed into animal feed as an additive. However, the EU have restricted this practice since 2002 due to the return of harmful substances to the human food chain [16].

In general, deep frying employs a larger amount of frying oil than pan frying, but both processes operate at a temperature range of 160–190 °C. The composition of the waste vegetable oil depends on several factors, such as the frying type, temperature, type of fried food and duration. Thus, the quality of the waste cooking oil fluctuates due to their different sources, including within and between house-holds, restaurants and food industries, but it mainly depends on the virgin vegetable oil used. In other words, the fatty acid profiles of waste vegetable oils change slightly from their parent oils.

The productivity of waste cooking oil in each country is unpredictable. However, it probably depends on many parameters, such as country size, population, food culture, food industries, and so on. For nearly the same country size, the USA produced 10 million tons of waste cooking oil, while the People's Republic of China generated 4.5 million tons of waste cooking oil in 2008. In the same year, Taiwan produced 0.07 million tons of waste cooking oil, which is approximately three-fold higher than that of Thailand, but Taiwan has half the population level and an over 10-fold smaller land area than Thailand. On the other hand, Japan and Malaysia generated approximately the same volume of waste cooking oil (0.5 million tons) yet Japan has a four-fold higher population than Malaysia with nearly the same area [17].

The selected properties and suggested testing methods for used vegetable oil sample are shown in Table 4. The composition of the waste vegetable oil is changed from that of the virgin oil by thermolytic (pyrolysis), hydrolytic (hydrolysis) and oxidative reactions [15]. These reactions are the source of the chemical contaminants which are measured by the increasing levels of FFA, water, total polar compounds, oxidized triglycerides, polymerized triglycerides, and the acid and peroxide values. Furthermore, the cracking of triglycerides and unsaturated fatty acids might lead to a decreased saponification number and iodine value, respectively. In addition to the chemical contaminants are the physical contaminants (% solid portion) that come from food processing, such as burned food bits, paper and aluminum foil. The physical contaminants, such as the visible solid portion, can be easily removed by simple filtration; whereas, the chemical contaminants require specific methods for their removal such as the acid esterification of FFA.

Regardless of the source of the waste vegetable oil, the major contaminants in the used cooking oils that affect the biofuel processing are the FFA and moisture content, which have been reported to vary within a wide range of 2–50% (w/w) and 0.5–20% (w/w), respectively [17]. To use the waste cooking oil as a biodiesel production feedstock, a suitable pre-treatment processes, such as the separation of suspended solids and removal of soluble salts, moisture and FFA, is essential, especially when employing the conventional homogeneous catalytic process. Although it has been reported that thermo-chemical processes, such as pyrolysis or catalytic cracking and supercritical techniques, are capable of dealing with these contaminants (including water and FFA) in waste cooking oils at a laboratory scale [18,19], they are not yet commercially viable.

3.2. Animal fats

Several animal fats, mostly obtained from animal slaughter and typically disposed in landfills, have been reported as potential alternative feedstocks for biofuels production. These include alligator fat [20], beef tallow [21], chicken fat [22,23], duck tallow [24], lamb meal [25] and other poultry fats [26]. The amount of animal fats which are produced in different areas depends on the food industries in those countries. For example, approximately 7500 t per year of alligator fat was produced in the southeastern United States. However, unlike waste cooking oils, information on the productivity of animal fats for other countries is presently deficient.

As with waste cooking oil, the major problem of using animal fat as a biodiesel feedstock in the conventional catalytic process is the high water and FFA contents in the animal fat. For example, alligator fat, beef tallow, chicken fat and pork lard have a FFA content of 8.0–11.0% (w/w), 3.6–15.0% (w/w), 5.0–25.0% (w/w) and 0.5–1.5% (w/w), respectively. Because the amount of FFA and moisture depends on various factors, such as storage conditions, storage time, animal fats container, humidity and temperature, the variation in the suitability of animal fats as a feedstock quality is very high and in the same range as for waste cooking oils. Furthermore, the effects of these factors have not been adequately investigated and reported in international publications.

According to the fatty acid profiles (Table 5), alligator fat, chicken fat and pork lard have a similar major fatty acid composition of

Table 4
The selected properties and testing methods for waste cooking oil samples.

Physical and chemical properties	Used cooking oil Sample			Testing method
	Canadian [99]	European [16,100]	Vietnamese [101,102]	
% Solid portion	19	20	N/R	Filtration/Centrifugal method
% Water content	7.3	1.1–1.4	N/R	Karl Fischer method/ ASTM E203
% Free fatty acid	1.4–5.6	2.4–3.1	0.4–1.2	AOCS Ca 5a-40
% Total polar compound	22	N/R	N/R	AOCS Cd20-91
% Oxidized triglycerides	4.72	N/R	N/R	IUPAC, 2.508 (1987)
% Polymerized triglycerides	1.43	N/R	N/R	AOCS Cd 22-91 or IUPAC, 2.508 (1987)
Acid value (mg KOH/g oil)	2.5–11.2	5.3–6.3	0.7–2.4	AOCS Te 1a-64
Saponification value (mg KOH/g oil)	177.87	204.3–195.1	0.272–264.1	AOCS Cd 3b-76
Density at40 °C (kg/m^3)	900	937–939	920	ASTM D1298
Peroxide value (m_{eq}/kg)	N/R	5.6–6.3	N/R	AOCS Cd 8b-90
Viscosity at 40 °C (cSt)	44.7	190.2–201.3	27.42–30.05	ASTM D445
Iodine value (g I_2/100 g oil)	N/R	104.3–115.3	8.57–13.2	AOCS Cd 1b-87

[a]N/R is not reported.

Table 5
Fatty acid profile of some commonly used animal fats.

Sample	Fatty acid content (% w/w)							
	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	Other
Alligator fat [20]	1.27	23.30	12.10	5.30	55.54			1.56 (C20:1) 0.28 (C20:3) 0.64 (C20:4)
Beef tallow [21]	2.69–4.00	22.30–28.00		19.20–41.28	28.82–48.90	0.58–3.00	0.50–2.00	
Chicken fat [22,23]		21.00–25.20	5.80–7.80	5.50–5.90	38.20–48.50	17.30–23.80	0.70–1.90	
Duck tallow [24]		17.00		4.00	59.40	19.60		
Lamb meal [25]/mutton fat [51]	4.0	10.10–28.10		6.00–27.20	35.00–31.28	1.59–36.00	0.88	2.00 (C20:0) 2.00 (C22:0)

Table 6
Fatty acid profile of some commonly used industrial wastes and by-products.

Sample	Fatty acid content (% w/w)					
	C16:0	C18:0	C18:1	C18:2	C18:3	Other
Yellow grease [30]	23.24	12.96	44.32	6.97	0.67	2.43 (C14:0)
Brown grease [30]	22.83	12.54	42.36	12.09	0.82	1.66 (C14:0)
Activated sludge [31]	28.00	7.50	24.90	9.50	2.00	18.00 (C16:1) 3.50 (C14:0) 7.50 (UNK)
Leather industry wastes [32] (12.2% FFA)	20.59–28.40	8.36–13.23	40.50–42.06	1.80–2.97	0.00–0.16	3.05–4.20 (C14:0) 4.60–8.10 (C16:1)
Waste fish oil [27] (4.90–10.7% FFA, 0.08–0.13% moisture)	27.30–28.10	11.70–12.10	42.80–44.60	9.40–10.50	0.50–0.70	4.00–4.50 (C16:1)
Tall oil [34,35]	1.00	3.00	60.00	32.00	2.00	1.00 (C20:3)
Rapeseed soap stock [37,38] (1.8% FFA)	4.80	2.90	87.60	0.6		
Palm soap stock [38]	(1.1% FFA)	47.00	4.50	42.90	0.2	1.20 (C14:0)
Olive soap stock [38] (1.3% FFA)	11.90	2.90	81.30	0.3		
Spent coffee grounds [40,41,103] (3.25–6.40% FFA)	43.65	6.49	8.15	32.4	1.31	3.57 (C12:0) 1.99 (C14:0) 2.39 (C20:0)
Citrus seed oil [42,104,105]	12.10–36.25	1.20–5.95	18.34–26.10	26.93–37.80	3.40–4.66	1.85–2.95 (C12:0)
Tomato seed oil [43]	12.26	5.51	22.17	56.12	2.77	

UNK is unknown, N/D is not detected and N/R is not reported.

palmitic and oleic acids, with minor fatty acids of myristic, stearic and linoleic acids. For duck tallow, it has up to 60% (w/w) of oleic acid (C18:1) and approximately 20% (w/w) of palmitic and linoleic acids, whilst beef tallow has the highest saturated fatty acid profile because it contains three major saturated fatty acids, palmitic (C16:0), stearic (C18:0) and oleic (C18:1) acids. In conclusion, the fatty acid profile of animal fats is slightly different from the edible and non-edible vegetable oils but does not significantly influence the biodiesel properties [11,12].

The acidic esterification pretreatment and either atmospheric or vacuum drying are usually applied to solve the high FFA and moisture content problems, respectively [27]. However, because of waste (especially waste water) management problems, heterogeneous catalysts have been applied in the pretreatment step instead [28]. For example, various metal precursors and supports have been evaluated as heterogeneous catalysts for biodiesel production from animal fat [28]. On the other hand, the supercritical alcohol technique was demonstrated to produce biofuel from chicken fat [22], lamb meal [25] and pork lard [26]. The catalytic cracking of beef tallow over activated charcoal supported palladium was also reported as an alternative method to produce biofuel [29].

4. Industrial wastes and by-products

Industrial wastes, mostly obtaining from waste water treatment, such as yellow grease (< 15% (w/w) of FFA) and brown grease (> 15% (w/w) of FFA) [30], activated sludge [31], leather industry wastes [32] and waste fish oil [27,33], have all been reported as potential additional feedstocks for biofuel production. Moreover, the industrial by-products, including the tall oil from the paper pulping process [34–36], soap stock for the fatty acid splitting process [37,38], spent coffee grounds from instant coffee factories [39–41], citrus seeds from orange juice production [42], and tomato seeds from ketchup and tomato paste manufacture [43], have all been reported as high potential feedstocks for biodiesel production. Formerly, those industrial wastes and by-products were worthless and were generally disposed of in landfills with the associated economic and environmental costs. Thus, the cost of these feedstocks is at present significantly lower than that of the edible and non-edible oils.

However, some industrial by-products, including citrus waste (seed and pulp) and tomato pomace (tomato skin and seed), have since been considered for livestock feed. A review on the productivity and availability of each industrial waste and by-product for individual countries and regions is presently lacking. Rather new industrial wastes and by-products are disclosed and reported in individual publications.

Palmitic (C16:0), oleic (C18:1) and linoleic (C18:2) acids are commonly found in industrial wastes and by-products (Table 6), and are also the major fatty acid components in palm oil. However, some fatty acids, such as palmitoleic (C16:1) and eicosatrienoic (C20:3) acids, which are rarely found in common plant oils, are found in some industrial wastes and by-products. The fatty acid profile of the soap stocks was observed to be similar as their parent oil. Even though the fatty acid profile of industrial wastes and by-products is somewhat dissimilar to the plant oils, it does

not significantly impact on the biofuel processing and fuel properties, as mentioned earlier.

It should be noticed that the crude tall oil is separated from the spent solution from Kraft's pulping process, commonly called the black liquor. Black liquor consists of various minor composition components that remain from the pulping process, such as lignin, inorganic salts, minerals and fiber. Due to the high amount of alkali in the black liquor, the principal component of crude tall oil is long chain carboxylic acid salts or soap. After acidification to promote phase separation, the tall oil soap converts to their parent FFAs and rosin acids. The FFAs in tall oil can be transformed to biodiesel (fatty acid alkyl esters), by esterification in the presence of an acid catalyst or under supercritical conditions [17,44]. However, rosin acids cannot be converted to biodiesel by this chemical reaction, but instead they can be transformed to biofuel via thermal processes, such as catalytic cracking [45]. The ratio between FFAs and rosin acids in the tall oil mainly depends on the source of wood. For instance, the North American softwood, the North American hardwood and Scandinavian pine consist of 47%, 76% and 50–55% FFAs and 42%, 0% and 30–35% of rosin acids, respectively. The tall oil yield was reported between 18 and 60 kg per ton of produced pulp [46].

5. Lipid-based biomasses from insects

Some insects, such as watermelon bugs (*Aspongopus viduatus*) [47], sorghum bugs (*Agonoscelis pubescens*) [47], black soldier fly larvae (*Hermetia illucens*) [48,49] oriental latrine fly larvae (*Chrysomya megacephala*) [50] and darkling beetle larvae (*Zophobas morio*) [51], have been reported as an alternative feedstock due to their moderate growth rate and high lipid content. For example, the total lipid content of the darkling beetle larvae, watermelon and sorghum bugs were reported as 33.8, 45.0 and 60.0% (w/w dry weight), respectively [47,51]. The oriental latrine fly larvae, raised in restaurant garbage for five days, had a lipid content of 24.4–26.3% (w/w dry weight). Furthermore, it was reported that the oil from the watermelon and sorghum bugs contain antioxidant compounds, such as tocopherols and sterols, that could improve the oxidation stability of the biofuel product.

The watermelon and sorghum bugs are pests of their respective plants, while the black soldier fly, darkling beetle and oriental latrine fly larvae are domestically grown as food for freshly hatched chickens, as well as food for reptiles and fish. Because of the simplicity of their life-cycle, larvae farming can be conducted at either a small scale using household equipment (e.g., plastic bucket and colander) or an industrial scale. The larvae can consume any waste containing cellulose (except wood), such as coffee pulp [52], proteinaceous waste like fish offal [53], and mixed organic substances, such as restaurant wastes [49] and organic leachates [54], a liquid fraction produced from vegetal and food waste, transforming this waste into a valuable lipid-based biomass. Thus, the oil derived from insects is a potentially promising lipid-based feedstock that avoids the economic competition between food and fuel industries and achieves the green conversion of waste to energy.

According to Table 7, the two major fatty acids of the oils from watermelon and sorghum bugs are palmitic (C16:0) and oleic (C18:1) acids, which is slightly different from that of palm oil. The oil derived from the black soldier fly larvae consists of the three major fatty acids of lauric (C12:0), palmitic (C16:0) and oleic (C18:1) acids. Due to the large amount of lauric acid in the oil from the black soldier fly larvae, the properties of the biodiesel derived from it might deviate slightly from that obtained from the oil of the watermelon and sorghum bugs. For example, the high saturated degree and a short carbon chain of lauric acid (C12:0) would be expected to make the resultant biofuel have a higher oxidative stability and a lower viscosity, respectively. However, the biodiesel obtained from black soldier fly larvae (97.2% methyl esters content) was reported to have a density, viscosity, flash point and Cetane index within the specific range of the international standard of biodiesel (EN14214) [48]. For oriental latrine fly and darkling beetle larvae, palmitic (C16:0), oleic (C18:0) and linoleic (C18:2) acids are their primary fatty acids and so are different from the fatty acid profile of the common oil plants.

In the conventional biodiesel production process, the FFA and moisture levels must be less than 0.5% and 0.1% (w/w), respectively [55]. The FFA content of watermelon and sorghum bugs' oil was higher at 3.0 ± 0.1 and 10.5 ± 0.1% (w/w) [47], respectively; whereas, the FFA content of the oriental latrine fly and Darkling beetle larvae were only 0.55 and 1.09% (w/w), respectively [50,51].

The moisture content for all insect oils shown in Table 7 was reported at less than 0.05% (w/w) but this is because the samples was dried and milled before being extracted by the organic solvent. In actuality, the drying, milling and extracting the lipid consumes a large amount of energy and so would add an additional economic cost and environmental impact. Against this, the combining of the extraction and reaction together into a single process as an in situ transesterification reaction has been demonstrated as a potentially viable alternative method [56].

6. Lipid-based biomasses from microorganisms

Oleaginous microorganisms are alternative lipid-based biomasses that have a high potential, especially in terms of their productivity. For instance, some oleaginous microorganism species accumulate lipid in their cell wall in range of 20–70% (w/w). After cultivation these microorganisms can reach a cell concentration of 0.1 kg per liter in a 1000 m^3 tank, from which approximately 3000 kg of lipid-based biomass could be obtained, which is higher than that from a 0.5 ha oil palm field [57].

Furthermore, genetic engineering to improve the productivity or the specific fatty acid composition can be more easily performed on

Table 7
The lipid content and fatty acid profile of selected insect oils.

Sample	Lipid content (% w/w)	Fatty acid content (% w/w)					
		C16:0	C18:0	C18:1	C18:2	C18:3	Other
Watermelon bug (*Aspongopus viduatus*) [47]	45.00	30.87–30.93	3.49–3.51	46.57–46.63	3.89–3.91	N/D	10.7 (C16:1) 2.4 (C17:0)
Sorghum bug (*Agonoscelis pubescens*) [47]	60.00	12.18–12.22	7.28–7.32	40.88–40.92	34.48–34.52	N/D	
Black soldier fly larva (*Hermetia illucens*) [48,49]	15.00–23.00	18.20–14.80	3.60–5.10	23.60–27.10	5.80–7.50	N/D	23.4–35.50 (C12:0)
Oriental latrine fly larva (*Chrysomya megacephala*) [50]	24.40–26.29	35.48	2.77	24.38	15.26	1.25	13.02 (C16:1)
Darkling beetle larva (*Zophobas morio*) [51]	33.80	32.74	9.36	29.43	22.53	0.85	2.16 (C16:1)

N/D is not detected.

many microbes (especially yeast and bacteria) than on insects (e.g., black soldier fly larvae) and oil plants (e.g., soybean and oil palm). In addition, the genetically engineered microbes can be completely contained (quarantined) inside the fermentation tank as this is a (theoretically) closed system in contrast to the open cultivation of insects or plants. Moreover, the cultivation conditions are easier to control, such as the temperature, nutrient types and levels, pH and, if applicable, light levels, and nor does the cultivating of oleaginous microorganisms require any insecticide, pesticide or herbicide. In addition, non-genetically modified oleaginous microorganisms can be cultivated in many different forms, such as cultivation in open ponds, raceways, photo-bioreactors or fermenters, allowing a greater flexibility of land usage. For example, an open pond requires a larger area than a fermenter does because its depth is limited by the unequal light intensity between the upper and lower levels of the pond.

The oil productivity of oleaginous microorganisms is commonly indicated as g/L/day or (kg/m^3/day) which differs from oil plants (kg/ha/yr), as shown in Tables 1 and 2. Therefore, the biomass and oil productivity of oleaginous microorganisms (Tables 8 and 10) are presented as kg/m^3/yr, based on 300 days of operation per year.

6.1. Microalgae

Some microalgae use photosynthesis to accumulate nutrients in lipid form, whilst others modify their metabolism in response to changes in the environment, such as nutrient or light levels, to synthesize and store lipid reserves. Accordingly, the growth rate and lipid content of microalgae depends on the growing conditions and several factors, such as light intensity, organic carbon and nitrogen sources, agitation speed, temperature, pH, salinity (for marine strains) and dissolved oxygen level. The growing

Table 8
The lipid content, biomass and oil productivity of selected microalgae.

Microalgae strain	Culture conditions	Lipid content (% w/w)	Productivity (kg/m^3/yr)	
			Biomass	Lipid
Chlorella sp. [74,106-108]	AT	22.4–33.9	158.4	53.7
Scenedesmus obliquus [64]	AT, MT	12.6–58.3	153	54.2
Pseudochlorococcum sp. [109]	AT	24.6–52.1	150	57.5
Chaetoceros muelleri [110]	AT	11.7–25.3	585	108.2
Tetraselmis chui [110]	AT	17.3–23.5	540	110.2
Chlorella zofingiensis [111]	HT	52	216	112.3
Crypthecodinium Cohnii [112]	HT	19.9	672	133.7
Nannochloropsis oculata [110]	AT	22.8–23	870	199.2
Chlorella protothecoides [113]	HT	48.1–63.8	412.5	230.8
Tetraselmis tetrathele [113]	AT	29.2–30.3	1125	334.7
Chaetoceros gracilis [113]	AT	15.5–60.3	1065	403.6
Schizochytrium mangrovei [114]	HT	68[a]	732	497.8
Schizochytrium limacinum [115]	HT	50.3[a]	1044	525.1

[a] As total fatty acid content. AT, MT and HT are autotrophic, mixotrophic and heterotrophic cultivations, respectively.

Table 10
The productivity of selected oleaginous yeasts, fungi and bacteria.

Yeasts or fungi strains	Lipid content (% w/w)	Productivity (kg/m^3/yr)	
		Biomass	Lipid
Yeasts			
Candida curvata [118]	29.2–58.0	690.6	315.0
Cryptococcus albidus [118]	33.0–43.8	251.7	146.0
Cryptococcus curvatus [118]	25.0–45.8	1989.7	1154
Lipomyces starkeyi [118]	61.5–68.0	635.7	410.0
Rhodosporidium toruloides [118]	58.0–68.1	3362.4	2120
Fungi			
Mortierella isabellina [10,119]	53.2	N/R	N/R
Mucor mucedo [10,119]	62.0	N/R	N/R
Aspergillus oryzae [76]	18.0–57.0	377.2	215.0
Cunninghamella echinulata [120,121]	35.0–57.7	232.2	134.0
Mortierella isabellina [120,122]	50.0–55.0	1275.8	678.8
Bacteria			
Arthrobacter sp. [118]	> 40	N/R	N/R
Acinetobacter calcoaceticus [118]	27–38	N/R	N/R
Rhodococcus opacus [118]	24–25	N/R	N/R
Bacillus alcalophilus [118]	18–24	N/R	N/R

N/R is not reported.

Table 9
The fatty acid profile of selected microalgae.

Microalgae strain	Fatty acid composition % (w/w)						
	C14:0	C16:0	C18:0	C18:1	C18:2	C18:3	Other
Chlorella sp [63,106]		6.7–19.1	1.1–4.4	8.5–9.1	2.4–14.4	15.5–18.8	5.24 (C15:0) 10.90 (C16:1) 0.3–13.8 (C16:2)
Chlorella zofingiensis [111]		22.6	2.09	35.7	18.5	7.75	1.97 (C16:1) 7.38 (C16:2) 1.94 (C16:3)
Pseudochlorococcum sp. [109]		53.0	N/R	42.0	N/R	N/R	N/R
Crypthecodinium cohnii [113]	13.4	22.9	2.6	7.9			2.9 (C12:0) 49.5 (C226)
Chaetoceros muelleri [116,117]	18.5–40.0	5.0–40.0	0.2–25.0	0.1–4.2	0.2–5.0	0.1–4.8	6.0–20.5 (C12:0) 0.1–8.0 (C16:2)
Scenedesmus obliquus [64]	1.48	21.8	0.45	17.9	21.7	3.76	5.95 (C16:1) 3.96 (C16:2) 0.68 (C16:3) 0.43 (C16:4)
Crypthecodinium cohnii [112]	13.4	22.9	2.6	7.6			0.4 (C16:1)
Schizochytrium mangrovei [114]		47.9–52.9	1.0–1.1	0.1		0.14	7.7–8.4 (C22:5) 28.7–30.7 (C22:6)
Schizochytrium limacinum [115]	3.3–4.0	53.6–60.5	1.34–3.9				4.5–6.5 (C22:5) 28.6–35.1 (C22:6)

N/R is not reported.

conditions that have been investigated in microalgae cultivations are phototrophic or autotrophic cultures (growth in light without additional nutrients), heterotrophic cultures (growth in dark with additional nutrients) and mixotrophic cultures (growth in either light or dark with additional nutrients). The lipid content, biomass and oil productivity of selected microalgae are summarized in Table 8.

From the lipid content in Table 8, cultivation in autotrophic conditions gives a higher fluctuation in the lipid content than that in heterotrophic conditions, which is probably due to the variation in the level of sunlight. The maximum oil productivity of *Schizochytrium limacinum* microalgae was 525.1 kg/m^3/yr, obtained by cultivation under a heterotrophic condition. Cultivation of *S. limacinum* in fermenters with a total volume of 10 m^3 yields similar lipid productivity as one hectare of oil palm.

The microalgae cultivation techniques can be categorized into the three main technologies of (i) an open raceway, (ii) a photo-bioreactor and (iii) a fermenter. Among the three operation modes, the raceway is the simplest with the smallest energy requirement and cultivation expense, but it can only be applied for autotrophic growth. An open-pond is easily contaminated with undesired microorganisms, such as bacteria or other microalgae species. Furthermore, cultivating in raceway yields the lowest cell concentration and, like that in an open pond, is dependent upon the variable abiotic (such as the weather including light intensity and CO_2 diffusion problems) and biotic (such as contamination with other organisms) factors.

For autotrophic cultivation, the use of a photo-bioreactor solves the weather dependence of the raceway, for instance, using fluorescent or light-emitting diodes (LEDs) lamps as a lighting source, and the contaminant problem by growing in a closed-system. The most common geometries of photo-bioreactor are a flat plate, annular or tubular configuration. Further details on the development of photo-bioreactors have been reviewed elsewhere [58,59]. On the other hand, some microalgae can be cultivated heterotrophically using organic substrates in the absence of sunlight, and here a fermenter can be used for their cultivation in mixotrophic or heterotrophic conditions [60]. Due to the readily available fermentation technology and knowledge base, the heterotrophic cultivation of microalgae is the one of the more promising technologies to produce lipid-based biomasses for biofuel production.

According to the fatty acid profile of selected microalgae strains, it is clear that lipids from microalgae have a more diversified fatty acid composition than plant oils (Table 9). In addition, some fatty acids, mostly polyunsaturated fatty acids (PUFAs), in microalgal lipids, such as hexadecadienoic (C16:2), hexadecatrienoic (C16:3), hexadecatetraenoic (C16:4) and eicosapentaenoic (C20:5) acids, are rarely found in plant oils. The presence of PUFAs affects the properties of the resultant biofuel, especially biodiesel, in terms of the viscosity, oxidation stability and cold flow properties. For example, the biodiesel obtained from microalgal lipids might have a lower viscosity and oxidation stability than the biodiesel produced from palm oil.

Despite the high lipid productivity of microalgae, the production cost of microalgal lipids is presently much higher than that of the plant oils. For instance, the price of microalgae-derived oil was reported to be approximately 5.00 US$/kg, which is some four- to five- fold higher than the prices of the common plant oils (Table 1) [61]. One of the major production costs of microalgae lipids is the harvesting step because of the small concentration of microalgae cells in the water, especially when cultivated in raceways. For industrial process, a low cell concentration of roughly 1–5% solids, is concentrated to 15–20% solids in the first step, and then to over 85% solids before the oil is extracted. Chemical flocculation combined with flotation or sedimentation and then removal of the water by centrifugation or filtration is the most promising cost and energy efficient method for concentration of the cells [62]. It should be noticed that lipid extraction methods at a laboratory scale, such as a one-step extraction-transesterification method [63] and rapid lipid extraction methods [64], are not realisable in a large scale extraction. Furthermore, the harvesting needs to be processed rapidly because the changing conditions during harvesting of the microalgae can induce cellular lipase induction and other metabolic changes leading to the rapid hydrolysis of the lipids to FFAs, which then pose negative effects on biodiesel production by the conventional method. In conclusion, besides improvement to the cultivating technologies, more sustainable harvesting and extracting technologies are also required. A review of downstream processing of microalgae is available elsewhere [65]. For example, an in *situ* reaction [66] and simultaneous extraction and conversion process [67] have been reported as shortcuts to produce biodiesel from microalgae.

6.2. Fungi and yeasts

Besides microalgae, many yeasts and fungi species are able to generate and accumulate lipids in their cell [68]. For example, several strains of yeast exhibit significant lipid production and accumulation levels with different substrates, such as sugar cane molasses [69], wheat straw [70] and rice straw hydrolysate [71]. Moreover, some oleaginous filamentous fungi (molds) can also produce lipids by utilizing the waste glycerol obtained from biodiesel production [72,73], sugar cane molasses [74], acetic acid, soluble starch, wheat straw, and wheat bran. The lipid content and productivity of selected oleaginous fungi and yeasts are presented in Table 10.

Unlike microalgae cultivation, fungi and yeasts are favorably grown in the absence of light, and so a fermenter is appropriate. Due to the advantages of cultivation in a fermenter (see Section 6.1), the yeast and fungal derived lipids are potentially promising lipid-based feedstocks. Some filamentous fungi produce lipids containing PUFAs, such as arachidonic acid (C20:4; ARA) and docosahexaenoic acid (C22:6; DHA) that are employed as infant food additives. Consequently, fungal lipids containing a high level of ARA or DHA are presently more expensive than most plant oils. Furthermore, the fuel properties of the resultant biofuel probably deviate from the biofuel obtained from the other feedstocks. The fatty acid profiles of lipids obtained from oleaginous yeasts and fungi are summarized in Table 11.

The fatty acid profile of the lipids from yeasts and fungi grown on selected media can be similar to that of the common plant oils, such as soybean and high oleic acid containing sunflower oils. It should be noticed that, however, the fatty acid profile of oleaginous yeasts and fungi significantly depend on the carbon source in the substrate. For example, variation in the fatty acid profile in the lipids from the yeast *Cryptococcus albidus* (see Table 11) was observed when different substrates (glucose and volatile fatty acids) were fed into the fermenter.

The carbon sources obtained from lignocellulosic or sugar-based biomasses are one of the most interesting substrates for yeast and fungi cultivation. The conversion of lignocellulosic biomasses to lipid-based biomasses by cultivation with oleaginous yeasts and/or fungi can utilize many agriculture, livestock, and food and household wastes as feedstocks for subsequent biodiesel production. When the hydrolysate of rice straw or wheat straw obtained from dilute H_2SO_4 pretreatment was investigated for use as a substrate for the oleaginous yeasts *T. fermentans* and *Aspergillus oryzae*, respectively, the lipid accumulation level was reduced in presence of acetic acid, furfural, 5-hydroxymethylfurfural and acid soluble lignin [71,75]. However, the removal (detoxification) of such

Table 11
The fatty acid profile of selected yeasty, fungal and bacterium lipids.

Yeasts and fungi strain	Fatty acid composition % (w/w)					
	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3
Yeast						
Candida curvata [119]	25.0–28.0		5.0–44.0	9.0–33.0	15.0–27.0	
Cryptococcus albidus [124]	16.1–17.9		5.1–0.5	2.9–17.7	19.6–61.1	0.5–59.1
Cryptococcus curvatus [125]	30.0–33.0		11.0–13.0	43.0–45.0	8.0–10.0	0.1–0.5
Lipomyces starkeyi [119]	36.2–37.1		4.5–5.5	45.1–46.3	3.4–4.9	
Rhodosporidium toruloides [120,126]	13.0–16.0		4.0–41.0	18.0- 42.0	15.0–29.0	
Fungi						
Cunninghamella echinulata [120]	17.7–30.1		5.8–11.3	52.1–56.3	4.2–12.4	
Aspergillus oryzae [74]	32.9		9.96	22.6	27.7	
Mortierella isabellina [123]	19.1–22.2	3.0	2.0–4.0	47.0–53.2	17.0–19.1	3.1–5.0
Bacteria						
Rhodococus opacus [76]	16.8–25.7		3.5–18.8	6.4–73.8	N/R	N/R

N/R is not reported.

from the wheat straw hydrolysate enhanced the lipid production by *A. oryzae* [75].

Moreover, some fungi species, such as members from *Phomopsis, Microsphaeropsis, Ephalosporium, Sclerocystis* and *Nigrospora*, simultaneously accumulate lipid and produce cellulase [76]. Due to the presence of cellulase, those fungi are able to digest amorphous cellulose in the lignocellulosic biomass to glucose and then anabolise this into a lipid-based biomass. Therefore, oleaginous yeasts and fungi could be a possible link between biofuel production from lipid-based and sugar-based biomasses.

6.3. Bacteria

Because of their very fast growth rate and simplicity, bacteria are of increasing interest as potential candidates for lipid-based biomass production. The lipid content of selected oleaginous bacteria is illustrated in Table 10. Although the biomass and lipid productivity of most of these bacteria are not available in the literature, it can be deduced that the productivity of bacteria is likely to be close to that of other oleaginous microorganisms due to their high lipid content in the dry cell.

With respect to the diversity of oleaginous bacteria, only a few bacteria can produce a high amount of the desired lipid type (triglyceride) in their cell. Rather, the lipids obtained from most bacteria are principally comprised of lipid complexes. For example, over 50% of the lipid extracted from *Arthrobacter globiformis* and *Arthrobacter scleromae* was comprised of glycolipids with a triglyceride content of only 1.9–25% (w/w) [77]. The complex lipids commonly accumulate in the cell membrane and are hard to completely extract by the traditional methods, such as cold press or solvent extraction. However, supercritical fluid extraction may be applicable to extract the oil from bacteria, since it has been established as suitable for lipid extraction from microalgae [78] and yeasts [79].

Due to their relatively new status as a potential feedstock for biofuel production, research on the fatty acid profile of bacterial lipids is currently lacking. However, as an example the fatty acid profile of lipids obtained from *Rhodococcus opacus* is illustrated in Table 11. When cultivated in a simple carbon source, such as gluconate or octadecane [77], the fatty acid profile of the lipids from *R. opacus* are similar to that of crude palm oil. Therefore, the carbon source for bacteria cultivation should be carefully selected to obtain a resultant biofuel which has fatty acid profile similar to that of the common plant oils. Moreover, the unusual fatty acids, such as phenyldecanoic acid and 4,8,12 trimethyl tridecanoic acid, were observed in *R. opacus* and *Nocardia globerula*, respectively, when cultivated on phenyldecane and 2,6,10,14-tetramethylpentadecanoic acid, respectively, as a carbon source [77]. The distribution of fatty acids in the bacterial oil is significantly dependent on the carbon source, as also observed for yeast and fungi. Due to the complexity of bacterial lipids, the thermal processes that aim to produce an alternative fuel, not only fatty acid alkyl esters (biodiesel), are more suitable than the transesterification process.

7. Recommendation and perspective of biofuels processing for the 2nd generation lipid-based feedstocks

To use the 2nd generation feedstocks in a conventional catalytic process requires a pre-treatment step to reduce the FFA and moisture contents to below a certain level (typically $<0.05\%$ and $<0.1\%$ (v/v), respectively). The pretreatment step generates additional costs and wastes that increase the economic and environmental impacts of using these feedstocks. The novel catalytic technologies, such as heterogeneous and enzymatic (lipase) catalysts, are an alternative route for dealing with the 2nd generation feedstocks [80,81]. Otherwise, processing techniques that are less sensitive to a high FFA and moisture contents could be employed. In addition, the thermo-chemical processes have more potential to integrate with the petroleum refinery because their operating parameters are nearly the same.

The systematic monitoring and control of the quality of the 2nd generation lipid-based feedstocks is particularly vital to their use in biofuel production. For instance, a complete set of procedures for the quality control of biodiesel feedstocks have been established by the American Oil Chemists' Society (AOCS), referral code AOCS Ck 1-07, including 23 AOCS standard testing methods. These testing methods consist of the testing of cleanliness, purity, water content, acidity, sulfur content, phosphorus content and oxidative stability. For other institutes, such as the American Society for Testing and Materials (ASTM) and International Union of Pure and Applied Chemistry (IUPAC), some testing methods for feedstock quality monitoring have been demonstrated, as illustrated in Table 4.

Even though considerable research on alternative feedstocks have been summarized in this article, it could not be clearly justified what is the most appropriate lipid-based feedstock for each community. For example, the cultivation of non-edible oil plants are suitable for some countries that have voluminous land and man power, whilst the oleaginous microorganisms are more suitable for some counties that have advanced fermentation and microbial technologies. The lipid-based biomasses obtained from industrial wastes and by-products could be employed as assisting feedstocks when the feedstocks from non-edible oil plants or

oleaginous microorganism are in shortage. Therefore, any biofuel production process that is able to employ a variety of different types and qualities of feedstocks is also an important strong point.

8. Conclusions

Besides the biofuel processing technologies, the source of lipid-based biomasses significantly influences the price of the resultant biofuel. The economic compatibility of biofuels derived from lipid-based biomasses is largely dictated by their price. It is clear that food-grade oil plants, the 1st generation feedstocks, are not appropriate feedstocks to produce inexpensive biofuel because their price is raised as a result of economic competition. The waste cooking oils, industrial wastes and by-products have a lower price than the 1st generation feedstocks, especially for the wastes that were formerly landfilled. Nonetheless, the price of the biofuel derived from these wastes is progressively increased by the complexity and number of downstream process, such as collecting, drying, handling and pre-treatment steps. On the other hand, insect and microorganism lipids have the advantage of a high productivity and a fast growth rate. Compared with edible and non-edible oil plant farming, the independence of climate provides a remarkable advantage for the cultivation of insects and microorganisms in a closed system. However, currently insect and microorganism derived lipids are more expensive than plant oils because they are currently not mature well developed technologies. In conclusion, research to minimize the 2nd generation feedstock price and processing costs, especially the downstream processing, are essentially required to obtain sustainable lipid-based biomass resources.

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